DESCRIPTION OF THE MORPHOLOGICAL AND TECHNOLOGICAL CHARACTERISTICS OF THE PLANT GOJI BERRY

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Abstract

Goji berry is relatively a new plant introduced in the Republic Macedonia. It is prevalent on limited small plantations to a controlled breeding conditions and providing a yield which is immediately placing on the market and awakens interest in further expansion of its production.This plant (Lycium barbarum or Lycium chinense), belongs to the family Solanacea and characterized by very favorable chemical composition of the fruit with high nutritional, energy and antioxidant value. Therefore, this plant is a large application in dietary recommendations for the prevention of many diseases and as a natural organic product for general strengthening of the human organism. From another aspect, goji berry requires minimal growing conditions aimed at irrigation and treatment with pesticides and it belongs in adaptable resistant plants for organic production. Also, this plant has good genetic predispositions, easily reproduced and has favorable characteristics; cytogenetic status, germination of seeds, regenerative ability, ability to clonal selection and in vitro - propagation. Its morphological construction is easy for examination of scientific purposes. Considering that in the fruit of this plant has similar chemical substances as it has in the grape, but enormously more, in our research experience made by fermentation (actually vinification) of the two types of Goji berries - sweet and bitter. The goal was to get fermented products similar to wine, but with improved chemical composition and strong antioxidant power.But after analyzing the obtained product, it can be concluded that by increasing the quantity of antioxidant substances during the vinification, however sensory evaluation of the final product is low. Goji berry should be consumed in fresh or dried form or may be a supplement of some special wines (with high sensory evaluation and strong antioxidant substances).

Key words: Goji, cytogenetic, antioxidant, germination, fermentation.

Introduction

The purpose of this paper is to examine in detail the cytogenetic status, the number of chromosomes, the properties of the seed material, the fertilization and reproduction of the goji berry plant as the genetic basis for its future selection and the utilization of its positive properties [8]. According to our examination in this paper, which is one of the initial studies of this type for this culture, it can be said that from the cytogenetic aspect, this plant has a diploid number of chromosomes with 2n = 24 and with few chromosomal irregularities, stable cell (mythological) divisions properly formed a partial spindle in the meiosis of the flowers, and thus a good fertilization of the flower [4]. The seeds have good germination and easily observed many correct mitotic divisions. The chromosome lengths are also measured and statistically processed. There is no frequent occurrence of triploidy and tetraploidy [2], [3]. The variation (division) of sweet and bitter fruits from the goji is not at the level of cell divisions and variations in chromosomes, but is due within the gene and part of the genomic structure. It could be the goal of future research on genomes and the germline dispersion according to NCBI matrices genomic maps for more families of plants worldwide [7], [9]. The fruit of goji can be used in fresh form, to dry, to get various drinks, juices, marmalades and fermented products, and from the seeds to extract oils and other types of nutrients. According to the fact that in the processing and fermentation of grains from some fruit

crops and from grapevines, juices or fermented products that are richer with nutrients and antioxidants and fresh fruits are obtained, we decided in this paper to process goji berry in a fermented product – wine.

Material and methods

For germination of goji berries we used a method used to germinate grapevine seeds. For obtaining material for observation under a microscope, it is necessary prior germination of the seeds of the examined godji berry which is performed by keeping the seeds in isolated plates, slightly covered with water, alternating 6 hours on a temperature of -2^{o}C to -3^{o}C, in a refrigerator and on 25^{o}C in a thermostat or a room temperature next to some heater. After the germination of the seeds they were slightly dried and planted in pots filled with garden soil and bio-fertilizer. For examination of the mitosis at the godji berry, germinated roots 5-10 mm long were used [9], [7]. For the examination of pollen and cell division of meiosis, methods used were applied in the flowers of the grapevine, by seeding and germination of pollen in a 15% solution of sucrose and treatment with orthocene. For the processing of goji berry in wine, standard fermentation was used which was used to obtain red wines. In the obtained goji berry wines we measured the content of alcohol, total extract, density were measured by picnometry. The amound of residual sugar, total acids, volatile acids, free and total SO_2 were analysed by titrimetry and pH with pH-meter. All of this methods were performed in the oenological laboratory of Agricultural Institute - Skopje, according to OIV methods. The obtained results for analysis of total polyphenols were expressed in mg/l EGA (Equivalente Galic Acid). Statistical processing according (SPSS) program. The tests were conducted in 2016/2017 year.

Results and discussion

The Goji plant, in the health food program, is increasingly being imported and grown in R. Macedonia. Goji's fruits have a high health and nutritional role in human nutrition. The fruits contain carbohydrates and carotenoids. Carotene pigments are represented by beta-carotene, zeaxanthin, lutein, lycopene, cryptosanthin. It also contains proteins, minerals (calcium, phosphorus, magnesium, iron, zinc and selenium) and vitamins (C, riboflavin, nicotinic acid and thiamine). *Lycium Barbarum* (Goji Berry) plants are native to the Himalayas. They also are referred to as wolf berries. Goji berries are called "super fruits" because of their antioxidant, amino-acid, essential mineral and protein content. The species of Wolfberry are deciduous woody plants growing 1-3 m. *L. Chinense* is cultivated in southern China and tends to be somewhat shorter, while L. barbarum is cultivated in the north, primarily in the autonomous region of Ningxia Hui, and tends to be slightly higher. Flowers grow in groups of one to three in the leaf axes [1]. The calyx (eventually interrupted by thegrowing berry) consists of bell-shaped or tubular separators forming short, triangular lobes [9]. The flowers are hermaphrodite, with pale purple and pale pink color, the pestle is straight, and the anteries are folded, which sometimes has the possibility of fertilization. In the northern hemisphere it can bloom from July to September and is fertilized, and the fruits ripen from July to October. The fruit is elliptical with a diameter of 1-2 cm and has an orange-red color in technological maturity. They are filled with small seeds (10 - 60 yellow seeds) [8], [11]. Synonyms for goji are „Wolfberry“, „Marriage grapevine“, „Chinese desert thorn“ etc. Goggi berry plant is suitable for growing in organic production, because it requires a maximum of 2-3 pesticide treatments throughout the year [5]. According to the observation and counting of metaphase chromosomes in the mitosis of cells from the examined material, only the diploid number of chromosomes AABBCC 2n = 24 was found [2], [3]. The chromosome length ranges from 3.50 µm to 5.65 µm in sweet goji berry and 3.49 µm to 5.64 in the bitter gobbies, which is a completely insignificant difference. (Fig. 1- 12, Chart 1). From the chemical composition it can be seen that the fermented product - wine from goji berry in relation to the red wine Vranec has the following differences; much higher content of total extracts, slight increase in phenolic substances and pH value, increased specific weight, while alcohols and CO2 are similar. In bitter goji berry has a very low residual sugar. (Table 3). As can be noted, according to the content of the rest of the sugar, the fermentation of wine in the current year is

completed, but the specific weight is still high. According to past data regarding this type of fruit, we assumed that the higher value of specific weight is due to the greater presence of pectin in the grains of this plant [1], [10]. According to the sensory analysis, the wine obtained from a slimy Goji berry and has a bitter taste with a typical characteristic flavor of the family Solanaceae. Our recommendation is that this alcoholic beverage could be used as an additive in the production of special wines to improve their antioxidant power or special type liqueur drinks [10], [1].

Table 1. Chromosome constitutions in a normally diploid organism with *2n* = 24 chromosomes - Goji berry (labeled A, B, and C) in the basic set

Name Goji berry / sweet, Goji berry / bitter	Designation	Constitution	Number of chromosomes
Monoploid	*n*	ABC	12
Diploid	2*n*	AABBCC	24
Triploid	3*n*	AAABBBCCC	36
Tetraploid	4*n*	AAAABBBBCCCC	48
Monosomic	2*n* – 1	ABBCC	23
		AABCC	23
		AABBC	23
Trisomic	2*n* + 1	AAABBCC	25
		AABBBCC	25
		AABBCCC	25

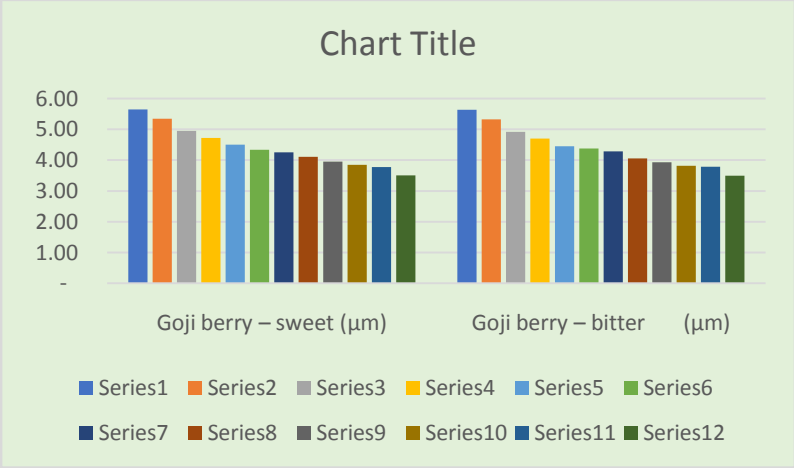

Chart 1. Graphical display of the length of the chromosomes

Table 2. Length of chromosomes in tested Goji berry and statistical processing

Haploid number of chromosomes N°	Goji berry – sweet (µm)	Goji berry – bitter (µm)
01	5,65	5,64
02	5,35	5,32
03	4,95	4,92
04	4,72	4,70
05	4,50	4,45
06	4,34	4,38
07	4,25	4,28
08	4,11	4,06
09	3,95	3,93
10	3,85	3,82
11	3,77	3,79
12	3,50	3,49
Average	4,41	4,40
*sd	0,65	0,65
*CV%	14,82	14,76
*L	5,65	5,64
*S	3,50	3,49
*L-S	2,15	2,15
*L+S	9,15	9,13
*L/S	1,61	1,62

*sd - standard deviation, *CV% - coefficient of variation, *L – longest chromosome,
*S – shortest chromosome, *L-S - difference of longest and shortest chromosome,
*L+S – sum of longest and shortest chromosome, *L/S - ratio between longest and shortest chromosome

Table 3. Chemical composition of fermented product - wine from goji berry and comparison with red wine Vranec

Parameters	Goji berry / sweet	Goji berry / bitter	Wine / Vranec
Specific weight 20/20	1,0137	1,0131	0,9983
Alcohol vol%	12,42	11,30	12,60
Total extract g/l	77,40	72,80	38,20
Residual sugar g/l	8,40	1,90	10,00
Total acids g/l	6,00	5,30	4,90
pH	4,63	4,91	3,45
Volatile acids lg/l	0,95	1,10	0,75
Free SO2 mg/l	15,36	12,80	26,88
Total SO2 mg/l	62,87	82,68	84,48
Total polyphenols mg/l	2569,60	2663,36	2400,00

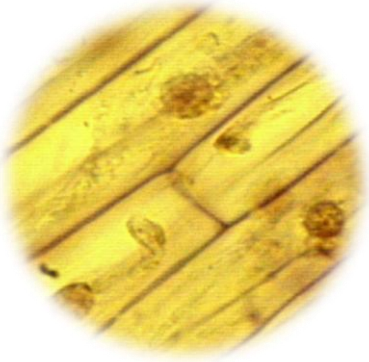

Fig. 1 Chromosomes of Goji berry – sweet

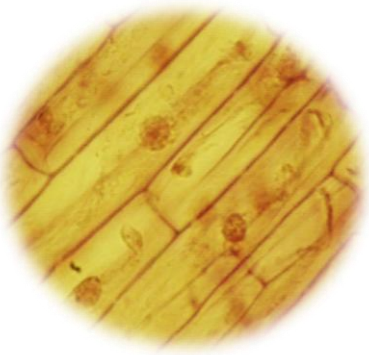

Fig. 2 Chromosomes of Goji berry – bitter

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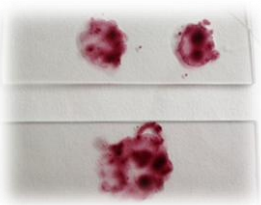

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Fig. 3 - 13 Grains, seeds of goji berry and processing of goji berry

Conclusions

Examined two types of goji berry - sweet and bitter showed to have uniformity regarding the number of chromosomes. The length of the chromosomes is within the cytogenetic properties of

the family Solanaceae and no major differences between the two variations sweet and bitter goji berry. In examined two types of goji berry, that sometimes may drastically reflect the cariotype and the polyploidia, no differences of that kind are noticedThe cell division is regular and the metaphase is normal. According to the existing limited possibilities for examination, abnormalities of the chromosomes are not noticed (aberrations, divisions etc.). According to chemical composition it is positive that the fermented product of the Goji berry - wine has a higher content of polyphenols that have a positive impact as nutritional components in food and beverages. After the sensory analysis and tasting, the final fermented product - Goji Beri wine was not rated with high marks. Therefore, this fermented product can be used as an addition to certain wines and some liqueur drinks to improve nutrition and health, ie. antioxidant value.

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References

Bennett, M.D., Price, H.J., Johnston, J.S., 2008. Anthocyanin inhibits propidium iodide DNA fluorescence in Euphorbia pulcherrima: implications for genome size vari- ation and flow cytometry. Ann. Bot. 101, 777–790.
Bernardello, G., Stiefkens, L., Las Penas, M.L., 2008. Karyotype studies in Grabowskia and Phrodus (Solanaceae). Plant Syst. Evol. 275, 265–269.
Jianjun Chen, Xiaoli Liu, Linyao Zhu, Ying Wang. 2013. Nuclear genome size estimation and karyotype analysis of Lycium species (Solanaceae). Scientia Horticulturae, 151, 46-50
Miller, J.S., Venable, D.L., 2002. The transition to gender dimorphism on an evolutionary background of self-incompatibility: an example from Lycium (Solanaceae). Am. J. Bot. 89, 1907–1915.
M.Vucinic, V. Pesic (1997) Manipulation of animal and plant genomes and genes in agriculture. Ed. Books, University of Belgrade
R. Dursun, Y. Zengin, E. Gündüz, M. Içer, H. M. Durgun, M. Dağgulli, I. Kaplan, U. Alabalik, C. Güloğlu, The protective effect of goji berry extract in ischemic reperfusion in testis torsion. *Int J ClinExp Med*, *8(2)*, 2015, 2727-2733.
Seo, G.S., Lee, J.Y., Kim, S.Y., Kim, J.K. & Han, G.H., 1986. The effects of fertilizer application level and top-dressing method on the yield component and fruit yield of Lycium chinense Mill. Korean Journal of Crop Science (Korea R.) 31(4): 465-469.
Stearn, W.T., 1972. Lycium L. In: Moore, D.M. (Editor): Solanaceae. Flora Europaea. Vol. 3. Cambridge University Press, United Kingdom. pp. 193-194
Yeung, K., Miller, J.S., Savage, A.E., Husband, B.C., Igic, B., Kohn, J.R., 2005. Association of ploidy and sexual system in Lycium californicum (Solanaceae). Evolution 59, 2048–2055.
Zhou, L., Leung, I., Tso, M.O., Lam, K.W., 1999. The identification of dipalmityl zeaxan-thin as the major carotenoid in Gou Qi Zi by high pressure liquid chromatography and mass spectrometry. J. Ocul. Pharmacol. Ther. 15, 557–565.
Zonneveld, B.J.M., 2010. Genome sizes in Hepatica Mill: (Ranunculaceae) show a loss of DNA, not a gain, in polyploids. J. Bot., http://dx.doi.org/10.1155/2010/758260
http://www.oiv.int
http://www.genres.de/eccdb/vitis
http://www.ecpgr.cgiar.org
https://www.ncbi.nlm.nih.gov/books/
http://www.vivc.de
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